ORIGINAL ARTICLE

Functionalization and Application of Carboxylated Multi-Walled Carbon Nanotubes for Adsorptive Removal of Penicillin G from Aqueous Solution

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Abstract

***Background*:** The presence of pharmaceutical contaminants such as Penicillin G in aquatic environments poses significant ecological and health risks. This study aimed to synthesize carboxyl-functionalized multi-walled carbon nanotubes (MWCNT-COOH) and evaluate their efficiency in removing Penicillin G from aqueous solutions through adsorption.
***Methods*:** This experimental study was conducted at the Toxicology and Pharmacology, Faculty of Pharmacy, Mazandaran University of Medical Sciences. MWCNT-COOH was synthesized via a diazonium-based surface modification and characterized using FTIR and SEM. Batch adsorption experiments were carried out to investigate the effects of pH, contact time, initial concentration, adsorbent dosage, and temperature. Adsorption data were fitted to Langmuir, Freundlich, and Temkin isotherms, and kinetics were assessed using pseudo-first-order and pseudo-second-order models.
***Results*:** The synthesized MWCNT-COOH showed efficient adsorption performance, with maximum removal observed at pH 8 and equilibrium achieved within 80 minutes. The Langmuir model best described the isotherm data, with a maximum monolayer adsorption capacity of 118.17 mg/g. Kinetic analysis revealed that the process followed a pseudo-second-order model, indicating chemisorption. Adsorption efficiency decreased with increasing temperature, suggesting an exothermic mechanism.
***Conclusion*:** MWCNT-COOH prepared via surface functionalization showed excellent potential as an adsorbent for Penicillin G removal. The findings support the use of functionalized carbon nanomaterials for the treatment of antibiotic-contaminated water.

Keywords: Penicillin G, Multi-walled carbon nanotubes (MWCNTs); Adsorption; Isotherm models; Kinetic modeling; Thermodynamic analysis; Wastewater treatment

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INTRODUCTION

The discharge of pharmaceutical compounds into aquatic ecosystems has emerged as a pervasive environmental challenge over the last two decades. These micropollutants originating from hospitals, pharmaceutical manufacturing, agricultural runoff, and domestic wastewater are often recalcitrant to conventional treatment processes and can persist at trace concentrations (µg/L) in surface waters, groundwater, and even drinking water supplies [1-3]. Among the wide range of pharmaceuticals detected, β lactam antibiotics such as Penicillin G (PG) are of particular concern due to their extensive use in both human medicine and livestock husbandry, as well as their potential to stimulate the development of antibiotic-resistant bacterial strains [4, 5].

Penicillin G is frequently detected in effluents from industrial and hospital wastewater treatment plants at concentrations up to several tens of µg/L, exceeding recommended environmental quality standards [6]. Its persistence arises from its chemical stability and partial biodegradability under typical biological treatment conditions, leading to continuous environmental exposure [7]. The presence of PG in natural waters has been linked to

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disruptions in microbial communities, toxicity to aquatic organisms, and horizontal gene transfer events that can propagate resistance genes across bacterial populations [8, 9].

Conventional wastewater treatment technologies—including activated sludge, membrane bioreactors, and advanced oxidation processes—offer limited removal efficiencies for Penicillin G. Biological treatments may transform the parent compound into metabolites that retain bioactivity or toxicity [10] ,while physicochemical methods such as chlorination and ozonation can generate harmful byproducts. Membrane filtration techniques provide higher removal rates but suffer from high operational costs, membrane fouling, and concentrate disposal challenges [11]. These shortcomings underscore the urgent need for alternative or supplementary strategies that are both effective and sustainable.

Adsorption has gained wide acceptance as a versatile and ecofriendly approach for the removal of pharmaceutical contaminants from water. Its advantages include simple operation, low energy demand, minimal formation of secondary pollutants, and the potential for adsorbent regeneration [8, 12[. A variety of adsorbents have been investigated, ranging from activated carbons and zeolites to biochars and polymeric resins, but many exhibit limited capacity or selectivity for antibiotics at environmentally relevant concentrations [13].

Carbon nanotubes (CNTs) have attracted significant attention in recent years due to their unique physicochemical properties: exceptionally high surface area (up to 1,000 m^2/g), tunable porosity, chemical stability, and the ability to engage in strong π–π and hydrophobic interactions with organic molecules [14]. In particular, multi walled carbon nanotubes (MWCNTs) offer advantages in terms of ease of production and cost, compared to single walled variants [4]. However, pristine MWCNTs tend to aggregate and exhibit low dispersibility in aqueous media, which limits their accessible surface area and adsorption performance [12].

To overcome these limitations, surface functionalization of CNTs with polar groups, especially carboxyl (–COOH) has been widely adopted. Carboxylation enhances hydrophilicity, introduces negative surface charge, and provides specific binding sites for cationic or polar organic pollutants via hydrogen bonding, electrostatic attractions, and π–π stacking [12, 15]. Several high-impact studies have demonstrated that COOH-functionalized MWCNTs exhibit superior adsorption capacities for dyes (up to 1,200 mg/g) and heavy metals (up to 500 mg/g), following Langmuir isotherm behavior and pseudo-second order kinetics [8, 16]. Yet, research focusing specifically on the adsorption of Penicillin G remains scarce.

Existing reports on antibiotic removal using CNTs primarily address fluoroquinolones (ciprofloxacin, norfloxacin) and tetracyclines, with only a handful of studies touching upon β lactams [17, 18]. These studies suggest that adsorption mechanisms for antibiotics can involve a combination of surface complexation, pore filling, and electrostatic interactions, but detailed investigations into the roles of pH, contact time, initial concentration, and temperature particularly for Penicillin G are lacking [19, 20].

Although various studies have investigated the adsorption of pharmaceutical contaminants using carbon-based nanomaterials, the removal of Penicillin G (PG), a widely used β-lactam antibiotic, remains significantly understudied, particularly with respect to carboxyl-functionalized multi-walled carbon nanotubes (MWCNT–COOH). Most existing research has focused on other antibiotic classes such as fluoroquinolones and tetracyclines, often neglecting β-lactams despite their frequent detection in wastewater and known ecological risks. Moreover, few studies have systematically examined the combined effects of operational parameters, including pH, initial concentration, adsorbent dose, and temperature, on PG adsorption performance using functionalized CNTs. Additionally, the adsorption mechanisms, kinetic models, and isotherm behaviors of PG on MWCNT–COOH have not been comprehensively explored. This gap in the literature underscores the need for detailed experimental investigations to evaluate the efficiency, capacity, and adsorption dynamics of COOH-functionalized MWCNTs in removing Penicillin G under varied environmental conditions.

The primary objective of this study is to evaluate the adsorption performance of carboxyl-functionalized multi-walled carbon nanotubes (MWCNT–COOH) for the efficient removal of Penicillin G (PG) from aqueous solutions under varying environmental conditions. To achieve this, MWCNTs were functionalized with carboxylic acid groups and characterized using FTIR and SEM to confirm successful surface modification. The effects of key operational parameters—including initial PG concentration, adsorbent dosage, solution pH, contact time, and temperature—on the adsorption efficiency were systematically examined through batch experiments. Furthermore, the adsorption data were analyzed using Langmuir, Freundlich, and Temkin isotherm models to elucidate the equilibrium behavior, while kinetic studies were performed using pseudo-first-order and pseudo-second-order models to determine the rate and mechanism of adsorption. The ultimate goal is to demonstrate the potential of MWCNT–COOH as an effective and reusable adsorbent for the removal of β-lactam antibiotics from contaminated water sources.

METHODS

Chemicals and Materials

This experimental study was conducted at the Toxicology and Pharmacology, Faculty of Pharmacy, Mazandaran University of Medical Sciences. The research protocol was reviewed and approved by the Ethics Committee of

Mazandaran University of Medical Sciences (Ethics Code: IR.MAZUMS.REC.1404.600).

All chemicals used in this study were of analytical reagent grade. Penicillin G potassium salt ($C_{16}H_{18}N_2O_4S \cdot K$) was obtained from Sigma-Aldrich (USA). Multi-walled carbon nanotubes (MWCNTs, purity > 95%, outer diameter 10–30 nm, length 10–30 μm) were purchased from Nanolab Inc. (USA). The reagents for functionalization, including 1,2-dichlorobenzoyl, 4-aminobenzonitrile, isopentyl nitrite, sodium hydroxide (NaOH), hydrochloric acid (HCl), acetonitrile, ethanol, dimethylformamide (DMF), and acetone, were procured from Merck (Germany) and used without further purification. Distilled deionized water was used throughout all experimental procedures.

Functionalization of MWCNTs with Carboxylic Acid Groups (MWCNT–COOH)

Functionalization of pristine MWCNTs was performed via diazotization followed by alkaline hydrolysis, based on modified literature protocols [21, 22]. Briefly, 0.024 g of dried MWCNTs was dispersed in 60 mL of 1,2-dichlorobenzene using ultrasonication for 15 minutes, followed by argon purging at room temperature. A solution of 4-aminobenzonitrile (3 g, 25 mmol) in 30 mL acetonitrile was added and stirred under an inert atmosphere. Isopentyl nitrite (4.5 mL, 35 mmol) was then introduced dropwise to initiate diazotization, and the mixture was maintained at 60 °C for 15 hours. After cooling, the product was washed, filtered (0.2 μm membrane), and subjected to a second reaction with 40% NaOH and ethanol (1:1 v/v) at 80 °C overnight. The suspension was acidified to pH 3 using HCl, filtered again, and the final MWCNT–COOH product was washed with ethyl acetate, water, and acetone, then dried at room temperature [21, 23].

Characterization of Adsorbent

Fourier-transform infrared spectroscopy (FTIR) was carried out using a Bruker Tensor 27 spectrometer to identify the surface functional groups before and after modification. Spectra were recorded in the range of 4000–400 cm^{-1} using KBr pellets [6].

Scanning Electron Microscopy (SEM) analysis was performed using a TESCAN MIRA3 instrument to study the morphological structure and surface characteristics of the CNTs before and after carboxylation. SEM images were captured at magnifications of 30,000× and 60,000× [24, 25].

Batch Adsorption Experiments

Batch adsorption experiments were conducted to investigate the removal of Penicillin G under varying conditions, including pH, initial PG concentration, adsorbent dosage, temperature, and contact time. Typically, 0.05 g of MWCNT–COOH was added to 20 mL of PG solution (10–50 mg/L) and placed in an ultrasonic bath at 298 K for 80 minutes. The pH was adjusted using 0.1 M NaOH or HCl before each test. After equilibrium was reached, the mixture was filtered, and the residual PG concentration was measured at 205 nm using a UV-Vis spectrophotometer (Shimadzu UV-1800). Each experiment was performed in triplicate, and the average values were reported [26, 27].

Adsorption Isotherm, Kinetic, and Thermodynamic Modeling

To evaluate the adsorption kinetics of Penicillin G onto MWCNT-COOH, batch experiments were conducted under varying initial concentrations, pH levels, temperatures, and adsorbent dosages. The amount of adsorbed antibiotic at any time t (q_t, mg/g) and at equilibrium (q_e, mg/g) was calculated, and the data were analyzed using pseudo-first-order and pseudo-second-order kinetic models [28].

The pseudo-first-order kinetic model, originally proposed by Lagergren, is expressed as follows:

$$\frac{dq_t}{dt} = K_1(q_e - q_t)$$

Its linear form is:

$$\log(q_e - q_t) = -\frac{K_1 t}{2.303} + \log q_e$$

q_e and q_t, respectively, are adsorbent adsorption capacities and adsorption absorption capacity at time t, the unit being both $\frac{mg}{g}$ and K_1 is the first-rate pseudo-velocity constant. To obtain the constant velocity and absorption capacity, the attractor of the curve log $\log(q_e - q_t)$ is plotted in t. Then, with the help of the gradient and the width of the origin of the curve equation, K_1 is obtained.

Pseudo-second-order model

This model assumes that chemisorption is the rate-limiting step:

$$\frac{dq_t}{dt} = K_2(q_e - q_t)^2$$

Its linear form is:

$$\frac{t}{q_t} = \left(\frac{1}{K_2 . q_e^2}\right) + \left(\frac{1}{q_e}\right) t$$

Where K_2 ($g \cdot mg^{-1} \cdot min^{-1}$) is the pseudo-second-order rate constant. A plot of $\frac{t}{q_t}$ Versus t was used to determine the kinetic parameters. The best-fitting model was selected based on the correlation coefficient (R^2) and agreement between experimental and calculated q_e values [29].

RESULTS

Characterization of the Functionalized Adsorbent

SEM Analysis

The morphology of the pristine and functionalized MWCNTs was examined using scanning electron microscopy. Unmodified MWCNTs displayed smooth tubular structures with minimal surface irregularities. After functionalization with carboxylic acid groups, the surface became more wrinkled and rougher, which is indicative of

successful chemical modification and increased surface activity (Figure 1).

FTIR Analysis

FTIR spectra further confirmed the introduction of

The effect of initial Penicillin G concentration (10–50 mg/L) on adsorption was studied under constant conditions (adsorbent dose: 0.05 g, pH: 9, temperature: 298 K). adsorption capacity increased with increasing

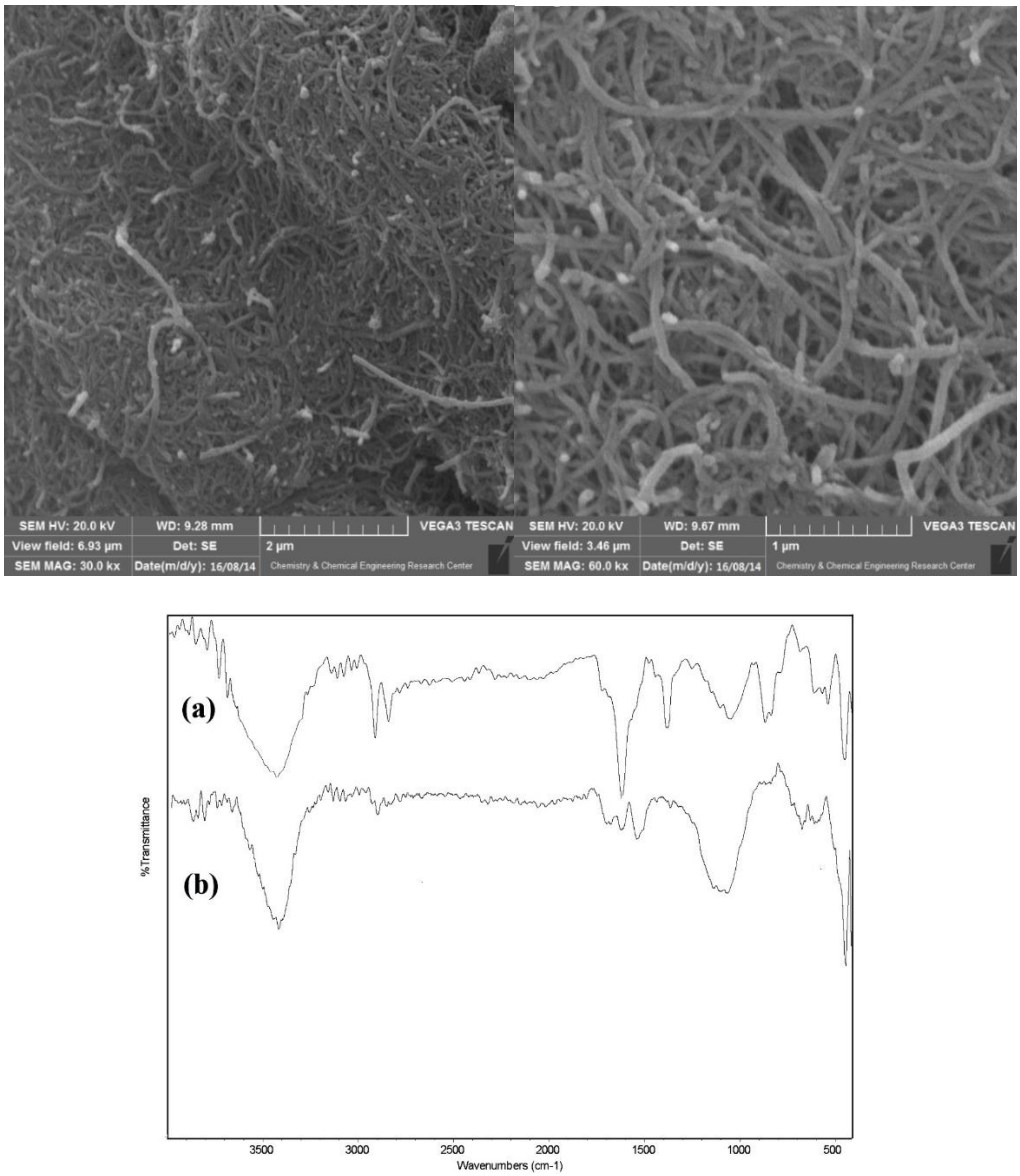

Figure 1. Structural and surface characterization of MWCNTs before and after functionalization: (a) SEM image of pristine MWCNTs (30,000× magnification); (b) SEM image of MWCNT–COOH (60,000× magnification); (a) FTIR spectrum of pristine MWCNTs; and (b) FTIR spectrum of functionalized MWCNT–COOH, indicating the appearance of hydroxyl and carbonyl functional groups following surface modification

carboxyl functional groups. strong absorption bands appeared at 3411 cm^{-1} (O–H stretching vibrations) and 1612 cm^{-1} (C=O stretching), which were absent or less pronounced in pristine MWCNTs. These findings confirm successful surface oxidation and the formation of MWCNT–COOH (Figure 1).

Effect of Operational Parameters

Effect of Initial PG Concentration

concentration due to a higher driving force for mass transfer and availability of more PG molecules to occupy adsorption sites (Figure 2a).

Effect of Adsorbent Dosage

Different MWCNT–COOH dosages (0.01–0.1 g) were tested while maintaining constant PG concentration (50 mg/L). Figure 4 shows that higher dosages improved adsorption due to increased surface area and availability of

active sites. However, excessive dosages may result in particle aggregation, reducing adsorption efficiency (Figure 2b).

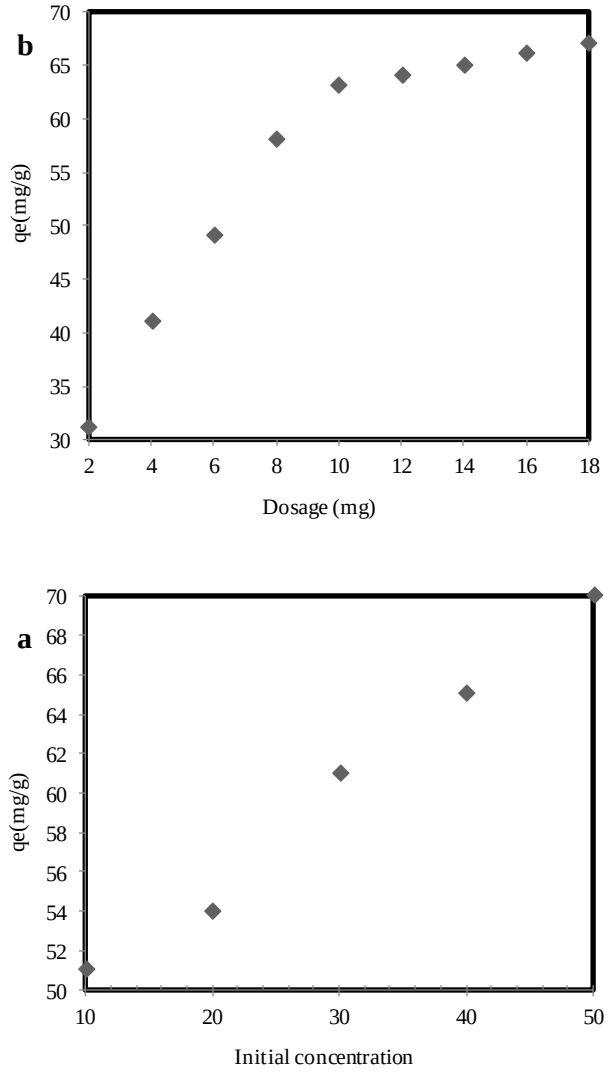

Figure 2. Effects of operational parameters on the adsorption capacity of MWCNT–COOH for Penicillin G: (a) initial Penicillin G concentration (range 10–50 mg/L; 0.05 g adsorbent, pH 9, 80 min, 298 K); and (b) adsorbent dosage (range: 0.01–0.1 g; 50 mg/L, pH 9, 80 min, 298 K)

Effect of Temperature

Adsorption studies were conducted at 298, 318, and 348 K. The adsorption capacity decreased with rising temperature, indicating an exothermic nature of the process. This suggests that lower temperatures are more favorable for efficient PG removal (Figure 3a).

Effect of pH

The solution pH (range 1–10) significantly affected the adsorption behavior. Figure 3b demonstrates that maximum adsorption occurred at pH 9, where the MWCNT–COOH

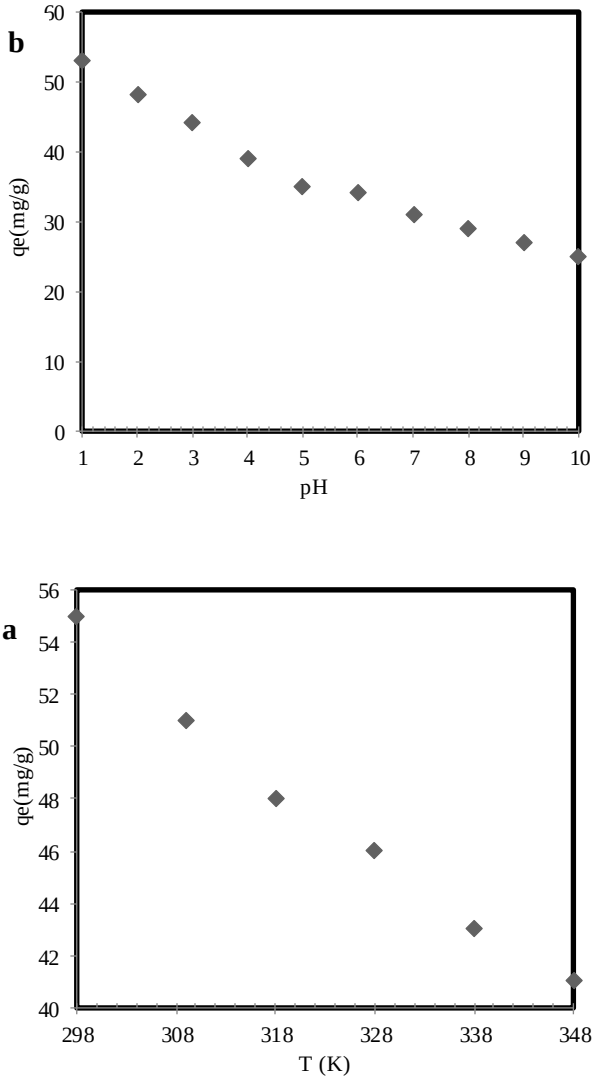

Figure 3. Effects of operational parameters on the adsorption capacity of MWCNT–COOH for Penicillin G: (a) temperature (298–348 K; 50 mg/L, 0.05 g adsorbent, pH 9, 80 min); (b) solution pH (1–10; 50 mg/L, 0.05 g adsorbent, 80 min, 298 K)

surface is negatively charged and favors electrostatic interactions with positively charged PG species. At low pH, proton competition hinders adsorption efficiency.

Effect of Contact Time

The equilibrium time was determined by varying the contact duration from 0 to 120 minutes. PG adsorption increased rapidly in the first 60 minutes and reached a

plateau at approximately 80 minutes, indicating that equilibrium had been achieved. This equilibrium time was used for all subsequent isotherm and kinetic experiments (Figure 4).

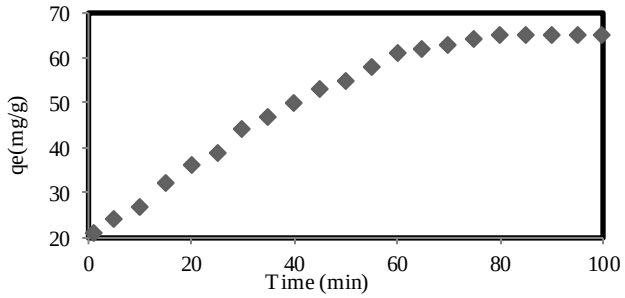

Figure 4. Effect of contact time (0–120 min) on the adsorption capacity of MWCNT–COOH for Penicillin G (50 mg/L, 0.05g adsorbent, pH = 9, 298 K)

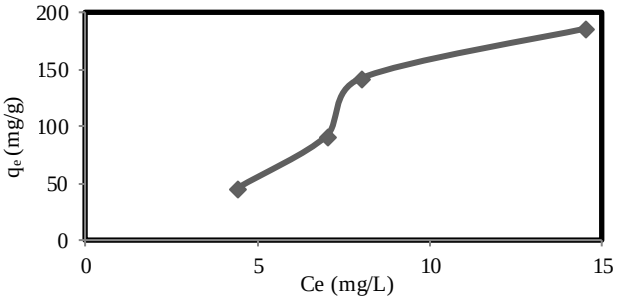

Figure 5. Adsorption isotherm models (Langmuir, Freundlich, and Temkin) for the surface uptake of Penicillin G (PG) onto MWCNT–COOH at 298 K and pH = 9

Adsorption Isotherms

To understand the adsorption mechanism, the data were fitted to Langmuir, Freundlich, and Temkin isotherm models. The Langmuir model provided the best linear fit with the highest correlation coefficient (R^2 = 0.998R), suggesting monolayer adsorption on a homogeneous surface. The maximum adsorption capacity (Q_0) was 118.41 mg/g. The Freundlich model also showed favorable conditions with K_F = 16.9, 1/n = 16.9/, and R^2 = 0.9821. Temkin constants suggested a moderate heat of adsorption. Full parameter values are summarized in Table1 and Figure 5.

kinetic parameters and correlation coefficients (R^2) are summarized in Table 1. The pseudo-first-order model yielded a calculated equilibrium adsorption capacity (q_e) of 112.5 mg/g and a rate constant (k_1) of 0.2125 min^{-1} with R^2 = 0.9321. In contrast, the pseudo-second-order model exhibited a higher correlation coefficient (R^2 = 0.9998), with a q_e value of 129 mg/g and a rate constant (k_2) of 0.053 g mg^{-1} min^{-1}.

DISCUSSION

This study investigated the adsorption behavior of Penicillin G (PG), a widely used β-lactam antibiotic, onto carboxyl-functionalized multi-walled carbon nanotubes (MWCNT–COOH) under various experimental conditions. The results demonstrated a high adsorption capacity of 118.41 mg/g, with optimal performance observed at pH 9, temperature 298 K, and contact time of 80 minutes. These findings confirm that MWCNT–COOH is an efficient adsorbent for removing PG from aqueous environments.

The significant enhancement in adsorption performance can be attributed to the surface functionalization of

Table 1. Adsorption modeling parameters for the removal of Penicillin G onto MWCNT–COOH, including isotherm (Langmuir, Freundlich, and Temkin) and kinetic (pseudo-first-order and pseudo-second-order) models. %ARE: average relative error; q_0: maximum monolayer adsorption capacity; K_L, K_F, and K_T: isotherm constants; k_1 and k_2: kinetic rate constants

Section A. Isotherm Models

Langmuir				Freundlich				Tempkin			
Q_0	K_1	R_1^2	%ARE	K_F	1/n	R_2^2	%ARE	K_t	B_l	R_1^2	%ARE
118.41	1.43	0.998	5.41	16.9	16.9	0.9821	8.01	1.71	16.9	0.936	612

Section B. Kinetic Models

Model	Parameters	R^2
Pseudo–first–order	q_e = 112.5 mg g^{-1} K_1 = 0.2125 min^{-1}	$R_1^2 = 0.9321$
Pseudo–second–order	q_e = 129 mg g^{-1} K_2 = 0.053 g mg^{-1} min^{-1}	$R_2^2 = 0.9998$

Adsorption Kinetics

To investigate the adsorption mechanism and rate-controlling steps of Penicillin G (PG) removal by MWCNT–COOH, the experimental data were fitted to both pseudo-first-order and pseudo-second-order kinetic models. The

MWCNTs with carboxylic groups (–COOH), which increases surface polarity, dispersibility in water, and the density of active binding sites. The negatively charged surface at alkaline pH enhances electrostatic interactions with the positively charged amino groups of the PG molecule, promoting higher adsorption efficiency. Similar trends have been reported in studies employing oxygen-functionalized carbon nanotubes and graphene derivatives for the removal of antibiotics such as tetracycline, amoxicillin, and ciprofloxacin [30-33].

The equilibrium adsorption data fitted the Langmuir isotherm model well (R_2 = 0.998), suggesting monolayer coverage of PG on a homogenous adsorbent surface. This observation aligns with studies showing that Langmuir behavior is dominant when strong specific interactions, such as hydrogen bonding or chemisorption, occur between adsorbate and adsorbent [34]. Additionally, the adsorption process followed pseudo-second-order kinetics (R_2 = 0.9998), indicating that the rate-limiting step is likely chemical adsorption involving electron transfer or covalent bonding. Similar kinetic profiles have been reported for the adsorption of antibiotics on oxidized nanomaterials and polymer-functionalized sorbents [35].

Compared to previous adsorbents used for antibiotic removal, including activated carbon, metal-organic frameworks (MOFs), and magnetic composites, MWCNT–COOH demonstrates competitive or superior adsorption capacity under mild conditions. For instance, Zhang et al. (2020) reported an adsorption capacity of 95 mg/g for tetracycline using GO-based composites, while functionalized CNTs achieved values in the range of 80–110 mg/g for sulfonamides and fluoroquinolones under similar conditions [33, 36]. The ease of surface modification, stability, and high surface area of MWCNT–COOH provide an advantage over more complex or cost-intensive materials.

One of the strengths of this work lies in its focus on Penicillin G, a relatively understudied antibiotic in adsorption research. Most prior [37] work has focused on tetracycline, ciprofloxacin, or sulfonamides, while β-lactams have received less attention due to their structural instability in aqueous media. By demonstrating high adsorption capacity for PG, this study addresses a critical research gap and suggests that carboxylated CNTs can be tailored to capture a broader spectrum of antibiotics.

Despite these promising results, there are some limitations. First, thermodynamic parameters were not evaluated, which would provide additional insight into the nature (endothermic vs. exothermic) and spontaneity of the adsorption process. Second, the regeneration and reuse of MWCNT–COOH were not studied. Since cost and sustainability are key in real-world applications, future studies should assess adsorbent reusability over multiple cycles. Third, although this study used model solutions, real wastewater contains multiple competing ions and organic compounds that can influence adsorption efficiency. Testing under realistic conditions will be necessary for practical implementation. In conclusion, this study demonstrates the potential of MWCNT–COOH as an effective, tunable, and relatively simple adsorbent for the removal of Penicillin G from aqueous solutions. Its high adsorption capacity, favorable kinetics, and functional group versatility make it a promising candidate for further development in wastewater treatment applications.

CONCLUSION

Carboxyl-functionalized multi-walled carbon nanotubes (MWCNT–COOH) demonstrated high efficiency for the removal of Penicillin G from aqueous solutions, achieving a maximum adsorption capacity of 118.41 mg/g under optimal conditions (pH 9, 298 K, 80 min). The adsorption process followed pseudo-second-order kinetics and was best described by the Langmuir isotherm model, indicating monolayer chemisorption on a homogeneous surface. Surface functionalization with –COOH groups significantly enhanced adsorption performance by improving polarity and electrostatic interactions. Overall, MWCNT–COOH represents a promising and effective adsorbent for antibiotic removal from contaminated water, although further studies on regeneration and real wastewater applications are recommended.

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Conflict of interest: None declared.

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